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Plasticisation of Carnauba Wax with Generally Recognised as Safe (GRAS) Additives

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Abstract

The plasticisation of carnauba wax has been examined by adding generally recognised as safe (GRAS) molecules of branched structures in order to disrupt the orderly packed molecules within the wax. The effectiveness of a range of different potential GRAS plasticisers was assessed by mechanical characterisation based on flexural and indentation measurements to determine parameters such as flexural strain and stress, fracture energy and Young's modulus. The microstructures of fracture surfaces were analysed using scanning electron microscopy (SEM) and the fracture surface roughness was measured utilising white light interferometry (WLI). The melting behaviour and crystallinity were investigated using differential scanning calorimetry (DSC) and x-ray diffraction (XRD). It is shown that polysorbates are effective GRAS grade plasticisers. The performance of all the materials investigated is rationalised in terms of the underlying molecular mechanisms of plasticisation, which contributes to the fundamental understanding of the plasticisation of wax.

Keywords

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Carnauba wax; Plasticisation; Hydrogen bonding.

1. Introduction

Carnauba wax originates from the Brazilian carnauba palm tree, formally known as *Copernicia prunifera*. This epicuticular wax has a wide range of applications in products such as confectioneries, polishing wax, paints, and cosmetics. One of its many useful applications is in dentistry, where carnauba wax can be used as an inlay casting composition and subsequently replaced by metal during casting.[1-3] However, it is brittle in the native state at room temperature and is the hardest naturally occurring commercial wax[4], which is a disadvantage for inlay casting of dentures that ideally requires sufficient plasticity so that they can be readily trimmed without tearing, chipping or flaking.[5, 6] Moreover, these characteristics are determined at room temperature since they dictate the ease with which trimming can be performed in a laboratory environment.[6] Hence, in practice there is a need to decrease the brittleness for ease of handling in order to facilitate carving. This demand for plasticisation may also be very important in other applications for which pliability is an important attribute. Nevertheless, the literature is sparse on the modification of carnauba wax to alter its mechanical properties for improving the ease of handling. Myers *et al.* filed a patent to plasticise a wide range of both natural and synthetic waxes using organic solvent-soluble salts of the general type R-O-C_nH_{2n}-COOH.[7] Nonetheless, the safety and toxicity of such salts are of concern for some applications and thus they would be unsuitable for many products that involve human or animal contact. The current paper describes the identification of some generally recognised as safe (GRAS) plasticisers that can significantly improve the pliability of carnauba wax.

Plasticisers can be categorised as internal or external. Internal plasticisers are co-polymerised into the final polymer structure and render the polymer molecules to be less ordered and more difficult to pack closely.[8] External plasticisers are low volatility molecules that produce swelling among the matrix molecules by means of their solvent ability without chemical reaction.[8] The widely accepted mechanism for polymer plasticisation is believed to be the

disruption of polymer-polymer interactions and the replacement with plasticiser-polymer interactions that increase the free volume within the polymer structure and so allows freer movement of the chains and hence an increase in flexibility.[9-12] le Roux reported that much of the structure and properties of Fischer-Tropsch waxes could be learnt from the closely related polyethylene polymer.[13] This may suggest that even though carnauba wax is a mixture of various molecules such as esters, hydrocarbons and fatty acids, the plasticisation mechanism for polymers could be transferable. Therefore the abovementioned mechanism is adopted for selecting potential plasticiser candidates. Since carnauba wax is lipophilic, the selected molecules should also be lipophilic or at least amphiphilic so that the interactions between the wax and plasticiser molecules are sufficiently strong. Moreover, it would be beneficial for the selected molecules to possess branch-structured groups in order to disrupt and inhibit the orderly packing and increase the free volume.

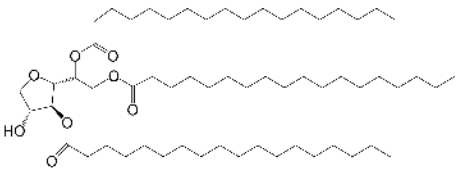
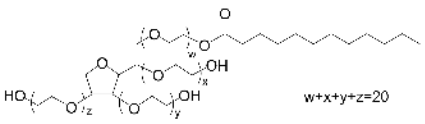
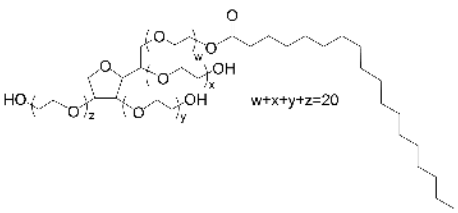
2. Materials and methods

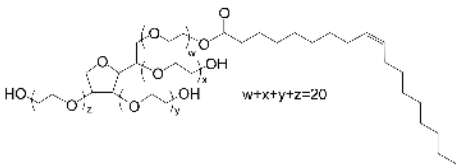
2.1. Materials

Based on the above discussion, Span®65, polysorbate 20, 60 and 80 were selected for potential plasticisation and some of their properties are listed in Table 1. Beeswax was also selected due to its unique plasticity.

Table 1 Physical and chemical properties of carnauba wax and the selected plasticiser candidates [14-18]

Candidate (acronym)	Formula	Molecular structure (Main composition)	Molar mass *M* (g/mol)	Appearance
Carnauba wax (CW)	Average chain length C_{50}	Aliphatic/aromatic esters (C_{44}-C_{66}) 84% (ω-hydroxy aliphatic esters 12-14%)	Average ~ 728	Solid

		Hydrocarbons (C_{16}-C_{24}) 2% Free fatty acids 3% Alcohols 3%		
Beeswax (BW)	Average chain length C_{40}	Esters 67% (monoesters (C_{32}-C_{52}) 35%, diesters (C_{56}-C_{66}) 14%, triesters (C_{72}-C_{80}) 3%, hydroxy polyesters (C_{72}-C_{96}) 8%, hydroxy monoesters (C_{40}-C_{48}) 4%, acid polyesters 2% (C_{40}-C_{50}) and acid monoester 1%) Hydrocarbons (C_{25}-C_{33}) 14% Free acids (C_{24}-C_{34}) 12% Free alcohols 1%	Average ~ 591	Solid
Span® 65 (SP65)	$C_{60}H_{114}O_8$		963.55	Solid
Polysorbate 20 (PS20)	$C_{58}H_{114}O_{26}$		1,227.54	Liquid
Polysorbate 60 (PS60)	$C_{64}H_{126}O_{26}$		1,311.70	Viscous liquid

Polysorbate 80 (PS80)	$C_{64}H_{124}O_{26}$		1,309.68	Liquid

Carnauba wax (CAS No. 8015-86-9, Henry Lamotte Oils GmbH), beeswax (CAS No. 8012-89-3, Koster Keunen) and Candelilla wax were kindly provided by DSM Nutritional Products Ltd (Switzerland). Span® 65 (sorbitan tristearate, Sigma 85547), polysorbate 20 (Sigma-Aldrich W291501), polysorbate 60 (SAFC W291609), polysorbate 80 (Sigma-Aldrich W291706) were all purchased from Sigma-Aldrich UK.

2.2. Specimen preparation for mechanical characterisation

An IKA C-MAG HS 7 heating plate was used to melt the carnauba wax or carnauba wax/plasticiser mixtures at 120 ℃. All mixtures were homogenised with an IKA Labortechnik Eurostar stirrer at 500 rpm for 5 min.

The specimens for flexural measurements were moulded into a cylindrical shape using 1 ml BD Plastipak Luer syringes with their tips cut off. To prevent poor flowability of the melted wax mixtures caused by solidification while cooling in contact with the syringes, they were heated to 120 ℃ in an oven before the moulding process. The homogenised melted mixtures were subsequently poured into the syringes and equilibrated at the laboratory temperature controlled at 17 ℃ to allow solidification. The moulded specimens were afterwards pushed out of the syringes and those with observable cracks were discarded. For each sample, 5 replicates were prepared. The diameter and length of the specimens were measured with a digital calliper (resolution 0.01 mm) to be 4.56 ±0.07 and 55.35 ±1.50 mm.

The specimens for indentation tests were moulded using glass petri dishes. Homogenised melted samples were poured into glass petri dishes and maintained at 17 ℃ until they solidified. The samples were then removed from the dishes. The diameter and height of

specimens were approximately 48 and 8 mm respectively. The lower surfaces were polished with grinding papers (grit size ×120, ×800 and ×2500) to eliminate the warping of the specimen when it was necessary. The central areas of the top surfaces were selected for the indentation tests.

2.3. Characterisation methods

2.3.1. Mechanical characterisation

Mechanical tests were performed on an Instron 5848 MicroTester with a 100 N load cell. The position resolution is 0.02 μm for speeds of < 200 mm/min. The actuator speed accuracy at zero or constant load is ±0.1% of the set speed. The accuracy of the load cell is 0.025 N when the load is ≤ 10 N, and 0.25% of the indicated load when it is > 10 N. An Instron 3-point bend configuration was used for the flexure tests. The radius of both the top and bottom anvils was 1.0 mm. The span between the lower two anvils was 30 mm. The loading speed was set to 0.05 mm/s. The compliance of the system including the flexure fixture tooling was measured to be 0.872 μm/N. For the indentation tests, a spherical glass indenter with a diameter of 6.35 mm was utilised and the loading speed was 0.01 mm/s. The compliance of the system including the tooling for the indentation test was measured to be 1.167 μm/N.

2.3.2. X-ray diffraction (XRD)

The specimens used for x-ray diffraction (XRD) tests were prepared following the same protocol. The glass petri dishes used as moulds had a diameter of approximately 40 mm. Aluminium foil was placed on top of a glass petri dish lid and the base was pressed into the lids to form a flat aluminium surface. This was to facilitate the mould removal, especially for the wax mixtures with polysorbates. The materials were weighed on a Sartorius Secura124-1S analytical balance with a resolution of 0.1 mg. A mass of 6 g homogenised melted sample was poured into the aluminium foil covered lid and the base was placed on top of the moulded specimen with its flat surface in contact with the sample after 90 s. Weights were placed in the petri dish base for 15 min in order to reduce warping of the specimen. The solidified specimen was then removed from the aluminium foil and cut with a disposable scalpel so that

it could fit in the Bruker D8 Advance XRD sample holder. The thicknesses of all the XRD specimens were measured to be 2.71±0.21mm. Blue tack was used underneath the specimens to adjust the height so that they were level with the holder edges. XRD diffractograms were recorded using a Bruker D8 Advance diffractometer with a LynxEye detector and Cu *K*α radiation restricted by a divergence slit 1.0 °. The x-ray diffractometer used did not have a monochromator. The data were collected using Cu *K*α1(1.54056 Å) and *K*α2 (1.54439 Å) radiation with *K*β (1.39222 Å) signals eliminated using a Ni filter. Peaks arising from Kα2 were subsequently stripped from the data using Bruker DIFFRAC.SUITE software. The scanning angle range was 15 °- 60 ° with an increment of 0.02 °. Samples were rotated at 30 rpm.

2.3.3. Differential scanning calorimetry (DSC)

A PerkinElmer differential scanning calorimeter 8000 was used for thermal analysis. The purge gas nitrogen flow was 20.0 ml/min. The reference sample was an empty Perkin-Elmer 40 µl aluminium crucible (Part No. BO14-3021) sealed with a PerkinElmer lid (Part No. BO143004). Each scanning was performed using 5-7 mg samples sealed in an aluminium crucible with a lid. The scanning programme was designed to hold for 5 min at 20 ℃ in order to equilibrate and subsequently commence data recording at a scanning rate of 5 ℃/min between 20 and 100 ℃. Three scannings were performed for each sample.

2.3.4. Scanning electron microscopy (SEM)

The surface morphology was investigated using a Hitachi TM3030 benchtop scanning electron microscope (SEM) with a back-scattered electron (BSE) detector. The observation condition and mode were set to energy dispersive x-ray (EDX) and charge-up reduction respectively. SEM micrographs were taken under a shadow imaging mode. All samples were coated with platinum using an EMSCOPE SC 500 sputter coater at 25 mA and a vacuum level of approximate 0.1 torr for 3 min before observation.

2.3.5. White light interferometry (WLI)

A KLA Tencor MicroXAM2 (supplied by Omniscan UK) optical interferometer with a ×50 Nikon lens was used for scanning the profiles of indents and studying surface roughness. The vertical and lateral scanning resolutions were 1 nm and 1 μm respectively. The specimens used for scanning indents were also coated with platinum to enhance light reflection.

3. Results and Discussion

3.1. Effect of plasticisers

3.1.1. Flexural loading characteristics

The effectiveness of the potential plasticisers was assessed by mechanical and physical measurements. The plasticity of materials is usually characterised by elongation using tensile tests. However, it is extremely difficult to prepare such specimens due to the physical properties of carnauba wax, such as its considerable brittleness and large volume contraction on cooling. Therefore, flexural and indentation tests were adopted to characterise the effectiveness of plasticisation.

Figure 1 illustrates the loading characteristics of pure carnauba wax and various mixtures with 30 w/w% of the selected plasticiser candidates. It can be clearly observed that pure carnauba wax possesses a brittle nature exhibiting a linear region and then an abrupt unstable fracture failure. Span® 65 only had the effect of slightly reducing the gradient of the loading curve and increasing the displacement at fracture. Beeswax had a similar effect although the gradient of the loading was further reduced and the displacement at fracture further increased. However, for polysorbates at the same concentration, the loading curves were non-linear with a smaller maximum failure force and a greater maximum failure displacement. In addition, failure was relatively stable for polysorbate 20 and 60 with no prominent crack propagation. This behaviour is characteristic of plastic deformation and clear evidence of effective plasticisation with the polysorbate 60 exhibiting the most pronounced effect.

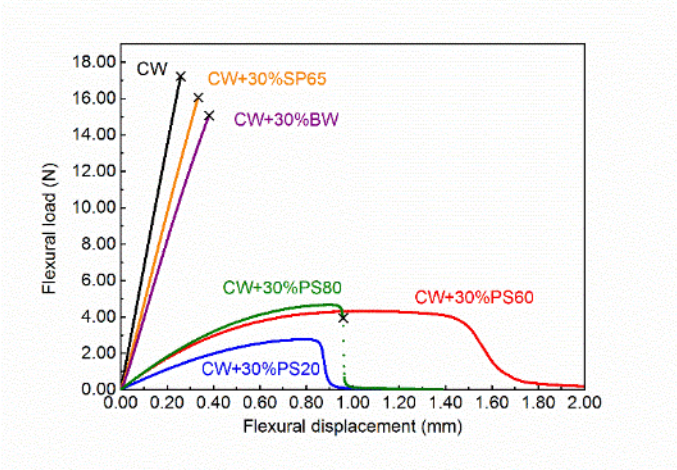


Figure 1 The loading characteristics of pure carnauba wax (CW), carnauba wax + 30 w/w% Span® 65 (CW+30%SP65), carnauba wax + 30 w/w% beeswax (CW+30%BW), carnauba wax + 30 w/w% polysorbate 20 (CW+30%PS20), carnauba wax + 30 w/w% polysorbate 60 (CW+30%PS60), and carnauba wax + 30 w/w% polysorbate 80 (CW+30%PS80)

The flexural stress for a circular cross-section beam during bending was calculated from the following expression:[19]

$$\sigma = \frac{DM}{2I} = \left(\frac{DFL}{4}\right) / \left(\frac{2\pi D^4}{64}\right) = \frac{8FL}{\pi D^3} \quad (1)$$

where D is the diameter of the circular cross section of the beam, M is the bending moment at the centre, I is the moment of inertia of the circular cross section, F is the load applied on the beam at the centre, L is the distance between the two constraining points. The flexural strain was obtained from:

$$\epsilon = \frac{6Dd}{L^2} \quad (2)$$

where d is the deflection of the beam at the centre. The ultimate flexural stress, *i.e.* the maximum flexural stress during bending, and the nominal flexural strain at fracture are presented as a histogram shown in Figure 2. The errors bars in the histograms hereafter represent 95% confidence limits of the mean values unless otherwise specified. Polysorbates

significantly reduce the ultimate flexural stress of the mixtures and increase the nominal flexural strains at fracture. Among polysorbate 20, 60 and 80, polysorbate 60 increases the flexural strain by a factor of ~ 6.5, the greatest compared with factors of 4.1 and 3.7 for polysorbate 20 and 80 respectively. The fracture energy, *G*, for each specimen was calculated from equation (3), where l_f is the displacement at fracture, and presented in Figure 3.

$$G = \frac{4}{\pi D^2} \int_0^{l_f} F \, dl \qquad (3)$$

The calculation of the fracture energy for the polysorbate 20 and 60 plasticised carnauba wax was achieved by defining fracture at cut-off force-displacement gradients of -10 and -5 N/mm , corresponding to a mean cut-off displacement of 1.06±0.22 and 1.66±0.29 mm respectively. The fracture energy has been increased by a factor of 2.6 for polysorbate 60, which has the greatest effect. The fracture energy of specimens plasticised by polysorbate 20 is of a similar order of magnitude to unplasticised carnauba wax due to the substantial reduction in the ultimate flexural load while the flexural displacement has been considerably elevated.

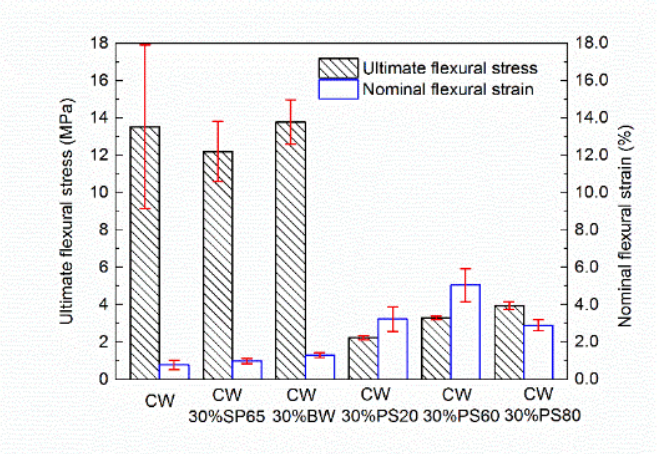


Figure 2 Ultimate flexural strength and nominal flexural strain at fracture for carnauba wax and various modified specimens

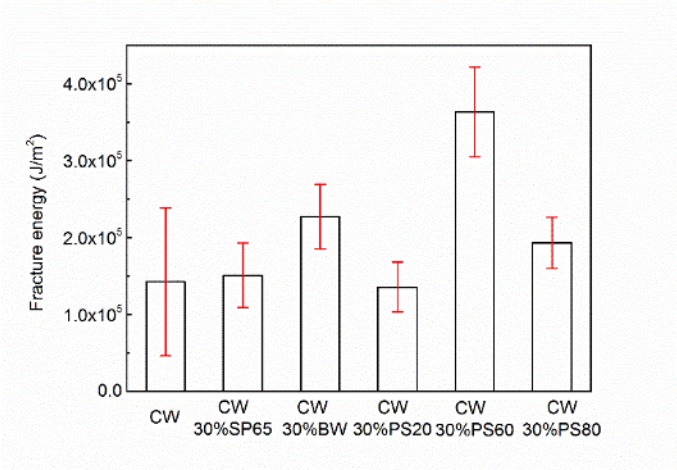


Figure 3 Fracture energy for carnauba wax and various modified specimens

3.1.2. Young's modulus

For the cases when the carnauba wax is plasticised, there was indentation at the support anvils during the flexural testing and this introduces uncertainty in the calculation of the Young's modulus (see 3.1.1.3). However, this parameter can also be obtained from the indentation tests utilising the Hertz equation as follows:[20]

$$F = \frac{4E\sqrt{R}}{3(1-v^2)}\delta^{1.5} \quad (4)$$

where F is load, E is the Young's modulus, R is the radius of the spherical indenter, v is the Poisson's ratio of the material tested and δ is the indentation depth. However the precise Poisson's ratio for various mixtures nonetheless is unknown and therefore a value of 0.5 was assumed. The values of the Young's moduli obtained from the two methods are shown in Figure 4 and the differences for each sample type are within experimental error. The large reduction in the moduli induced by the polysorbates clearly show that they are effective plasticising agents for this wax.

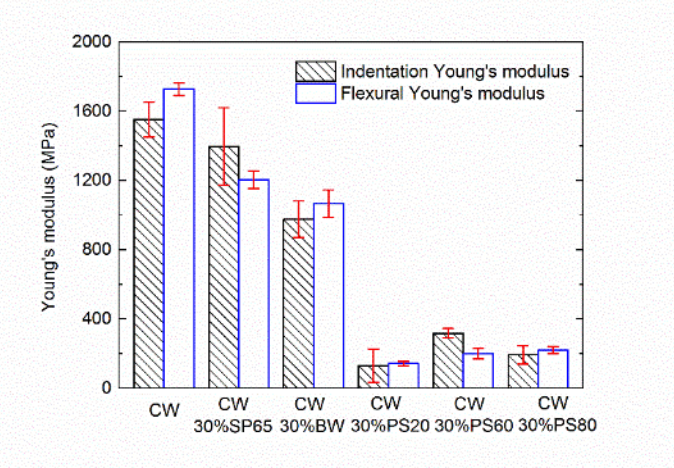


Figure 4 Young's moduli of various specimens calculated from both the flexural and indentation tests

It should be noted that there is a phase separation for the CW+30%PS20 mixture while on the hot plate when it was not stirred with the wax as the upper phase. No such phenomenon was observed for CW+30%PS60 and CW+30%PS80 mixtures. This composition inhomogeneity is less severe on a macro scale as shown in the flexural Young's modulus error bars in Figure 4, which proved that the solidification rate is rapid. However, the phase separation is likely to account for the much wider distribution of the indentation Young's modulus for CW+30%PS20 specimens as shown in Figure 4.

3.1.3. Indentation depth

The indents for the CW+30%PS20 and CW+30%PS60 cylindrical specimens for the flexural tests were scanned using an interferometer in order to assess the error that would be induced in the calculation of the Young's modulus. For each specimen, three upper and three lower indents were scanned. The sum of indentation depths for either specimen was << 80 μm, as shown in Figure 5. This indicates that the indentation on the specimens during flexural tests accounts for << 10% of the total strain.

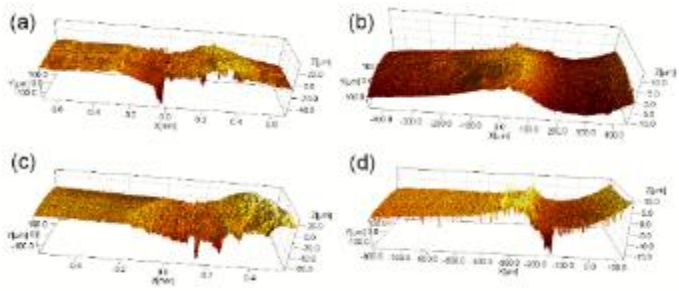


Figure 5 3D topography of indentation marks on the flexural test specimens scanned by an interferometer (a) bottom (b) top indents of a CW+30%PS20 specimen (c) bottom (d) top indents of a CW+30%PS60 specimen

3.1.4. Fracture surface roughness

The roughness parameter Ra of the fracture surfaces was scanned using an interferometer and the results are presented in Figure 6. Each scanning area was 127 × 171 μm. For pure carnauba wax, the mean roughness of the fracture surfaces is 1.37 μm. Mixing carnauba wax with 30 w/w% Span® 65 or beeswax increases the fracture surface roughness slightly to > 3.00 μm. The mean fracture surface roughness for polysorbate 20, 60 and 80 plasticised carnauba wax specimens is 6.74, 6.99 and 4.94 μm respectively. The surface is shiny for brittle fractures while dull for ductile fractures.[21] This is likely to be caused by the different light reflection conditions from these two types of fracture surfaces. Brittle fractures result in less rough surfaces than ductile fractures, leading to more light reflected and hence increased gloss. Therefore, the greater surface roughness of the fracture surfaces for the polysorbates is also likely characteristic of a plastic failure.

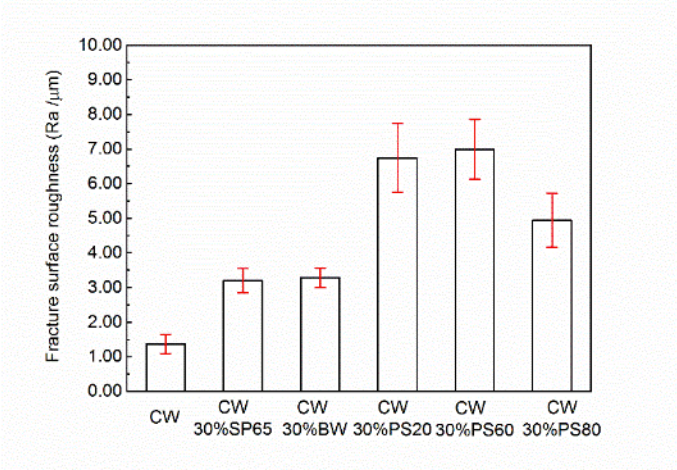


Figure 6 Fracture surface roughness (Ra) of carnauba wax and various modified specimens

3.1.5. Fracture surface morphology

The fracture surfaces of CW, CW+30%SP65 and CW+30%BW appear smooth and silky when viewed with the naked eye, whereas those of the polysorbates plasticised specimens are dull and dimpled as reflected in the surface roughness values described in the previous section. The microstructures were further observed using SEM and the micrographs are presented in Figure 7. Figure 7 (a) is a local region of the fracture surface of the pure carnauba wax specimen. Both the SEM micrograph and the interferometry result confirmed its smooth texture. Figure 7 (b) and (c) illustrate the fracture surface topography of specimens modified by Span®65 and beeswax. Neither is substantially different from pure carnauba wax, contrary to the more prominently spiky and flaky features presented by specimens plasticised by the polysorbates, as shown in Figure 7 (d_2), (e_2) and (f_2).

The microstructure of the fracture surfaces for CW+30%PS60 specimens appears to be consistent between regions, as well as between specimens, as shown in Figure 7 (e_1) - (e_3). The dissimilarity in microstructures between regions is more recognisable on a macroscopic scale for CW+30%PS80 specimens, as shown in Figure 7 (f_1). This could be because the fracture still has some brittle nature accompanying the plastic bending, as indicated by the terminal unstable fracture (Figure 1). No such prominent difference could be observed on the fracture surface for CW+30%PS20, as shown in Figure 7 (d_1). This is consistent with the

finding that this type of sample exhibits plastic hinging rather than crack propagation during failure (*cf*. Figure 1), and that the morphological inhomogeneity is less severe on a macro scale. However, on a microscopic scale, there are flaky and rough microstructures, as illustrated in Figure 7 (d_2); and less flaky and smooth microstructures, as shown in Figure 7 (d_3).

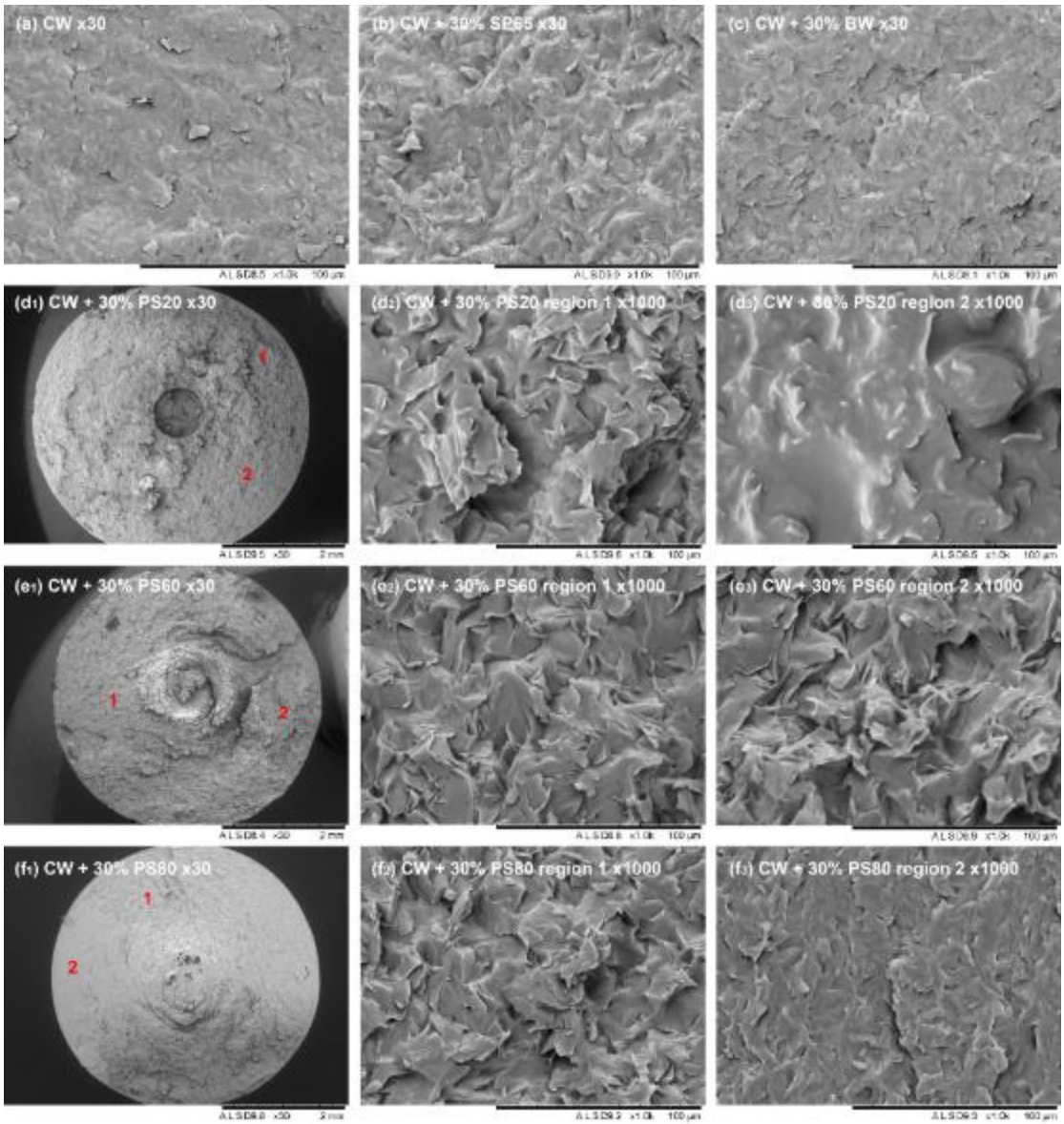


Figure 7 SEM micrographs of the fracture surfaces of pure carnauba wax and specimens modified by 30 w/w% various plasticisers

3.1.6. Melting point

Pure carnauba wax has a mean melting peak of 84.05±1.34 ℃ based on the three DSC measurements. Adding Span®65 and beeswax only slightly shifts the mean melting peak to 81.84±0.81 and 81.59±1.44 ℃ respectively. However, there is a small peak in the thermogram between 50 and 55 ℃ before the large peak for Span®65 plasticised sample as shown in Figure 8. This probably corresponds to a phase transition of the Span®65, which has a melting point of 53±3 ℃ approximately. The mixture of beeswax and carnauba wax exhibits a broad phase transition range, due mainly to the overlap of the melting ranges of both constituent waxes around 70 ℃. The average melting peaks for polysorbate 20, 60 and 80 mixed carnauba wax are 84.09±1.03, 85.23±2.10 and 85.19±0.54 ℃ respectively, which are very similar to the values for pure carnauba wax. In summary, the melting peaks appear to be similar for all mixtures.

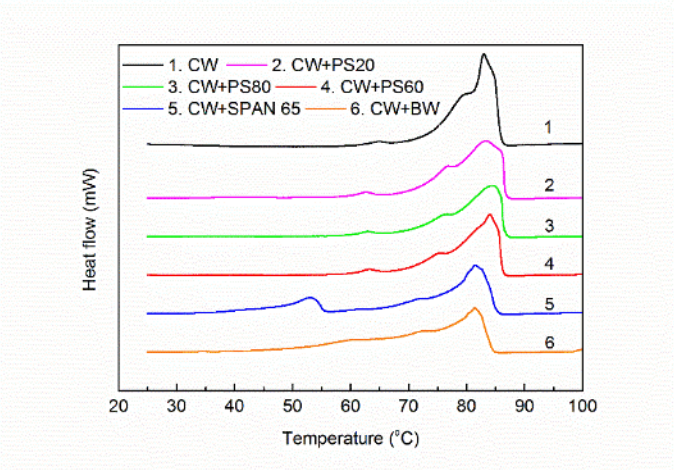


Figure 8 DSC thermograms of carnauba wax and various mixtures

3.1.7. Degree of crystallinity and crystallite size

Two large peaks were identified within the 2θ range of 20-22 ° and 22-24 ° corresponding to the (110) and (200) planes in an orthorhombic crystal system, as shown in Figure 9. The degree of crystallinity was calculated from equation (5):[22, 23]

$$X_c(\%) = \frac{A_c}{A_c + A_a} \times 100\% \quad (5)$$

where A_c is the area under the crystalline peaks and A_c+A_a is the global area including both crystalline (A_c) and amorphous areas (A_a).[24] The degree of crystallinity was obtained from the EVA software accompanying the diffractometer and the results are listed in Table 2. Beeswax slightly increases the degree of crystallinity of carnauba wax. Polysorbate 60 reduces the degree of crystallinity by approximately 10%, which is the most among the potential plasticisers examined.

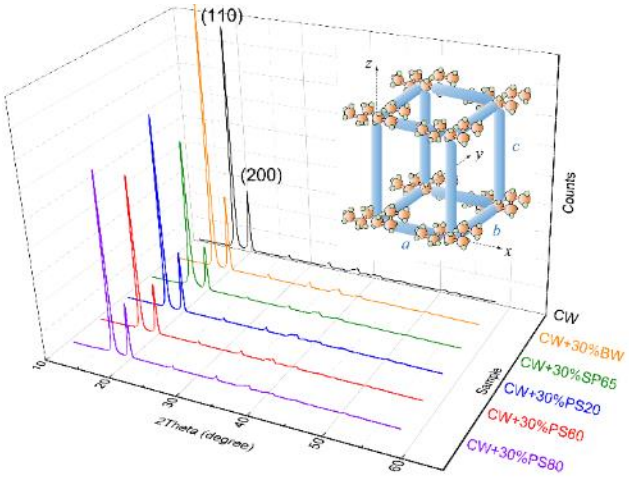


Figure 9 XRD diffractograms of carnauba wax and various mixtures scanned from a 2θ range between 15 ° and 60 °. The inset figure illustrates the orthorhombic crystal system ($a \neq b \neq c$) of carnauba wax. The wax molecules are for illustration purpose only and do not truly represent its molecular structure.

The crystallite size can be estimated by the Scherrer equation:[25]

$$L = \frac{K\lambda}{\beta \cos\theta} \quad (6)$$

where L is the crystallite size measured along the specific direction normal to the lattice plane given by the 2θ peak position, K is the Scherrer constant, λ is the wavelength of the x-ray, β is the peak width in radians. The full width at half maximum (FWHM) model was adopted to

determine the peak width, which defines β as $\Delta 2\theta$ at half of the peak maxima in equation (6); K was consequently taken to be 0.9 following the selection of the FWHM method.[26] The two large peaks were both used to estimate the crystallite size and the results are listed in Table 2. It should be noted that the Scherrer equation used is a simplified model that applies to an ideal system where a single peak is broadened due only to the crystalline size without sample defects or instrumental effects. In reality, non-zero contributions from other factors likely lead to an underestimation of the crystalline size. Nonetheless, to acquire the absolute crystalline size precisely is beyond the scope of the current research and it was only intended to compare the relative magnitudes of crystallite size.

Table 2 Degree of crystallinity and crystallite size for various samples

Specimen	(110) Peak			(200) Peak			Degree of Crystallinity
	Intensity (Counts)	FWHM (Rad)	Crystallite size (nm)	Intensity (Counts)	FWHM (Rad)	Crystallite size (nm)	
CW	218,182	0.0052	27	62,315	0.0062	23	68.3%
CW+30%BW	250,987	0.0052	27	74,107	0.0059	24	71.1%
CW+30%SP65	141,561	0.0066	22	43,546	0.0066	22	65.1%
CW+30%PS20	181,529	0.0041	34	57,353	0.0052	27	60.9%
CW+30%PS60	144,922	0.0048	29	47,761	0.0052	27	57.6%
CW+30%PS80	166,419	0.0045	31	50,848	0.0055	26	58.1%

3.2. Effect of polysorbate concentration

The mechanical measurements revealed that polysorbates are the most effective plasticisers of those investigated in the present work. In order to establish the effect of plasticiser concentration, further measurements were performed. The loading curves from flexural measurements and SEM micrographs of the fracture surfaces of specimens plasticised by 10 and 20 w/w% of polysorbate 20, 60 and 80 are presented in Figure 10 and

Figure 11. When the plasticiser concentration is 10 w/w%, the fracture surfaces of all specimens seem to be uniform, as shown in

Figure 11 (a_1) (c_1) and (e_1). This may be attributed to the fracture mode being mainly brittle and unstable. When the plasticiser concentration is increased to 20 w/w%, the fracture surface roughness starts varying from region to region, as shown in

Figure 11 (b_1)-(b_3), (d_1)-(d_3) and (f_1)-(f_3). This is probably caused by the appearance of a significant fraction of plastic behaviour. When the plasticiser concentration is further increased to 30 w/w%, the fracture surface roughness for polysorbate 20 and 60 modified specimens is greater than that of polysorbate 80, as confirmed by the histogram in Figure 12 and SEM micrographs in Figure 7. This suggests that the plasticising effect of polysorbate 80 is inferior to polysorbate 20 and 60, which is in agreement with the loading characteristics. As the concentration of plasticiser increases, the flexural stress, flexural modulus and fracture energy all decrease, suggesting that the mixtures become softer and weaker, as shown in Figure 13 (a), (c) and (d). However, the nominal flexural strain does not increase with concentration for polysorbate 20 and 80, as shown in Figure 13 (b). Therefore, there is no additional benefit to further increasing the concentration of these two plasticisers above 10 w/w% in terms of the failure strain as a key indicator of pliability although this is not the case for polysorbate 60. The fracture energy values of 20 and 30 w/w% polysorbate 60 modified carnauba wax are approximately equal.

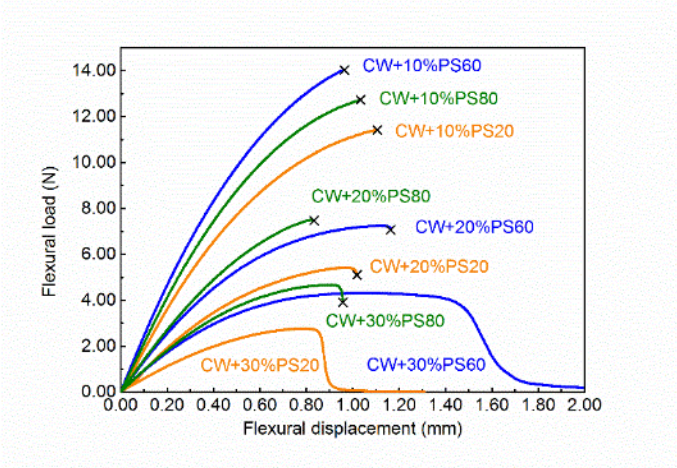


Figure 10 The loading characteristics of carnauba wax (CW) plasticised by different concentrations of PS20, PS60 and PS80 in weight

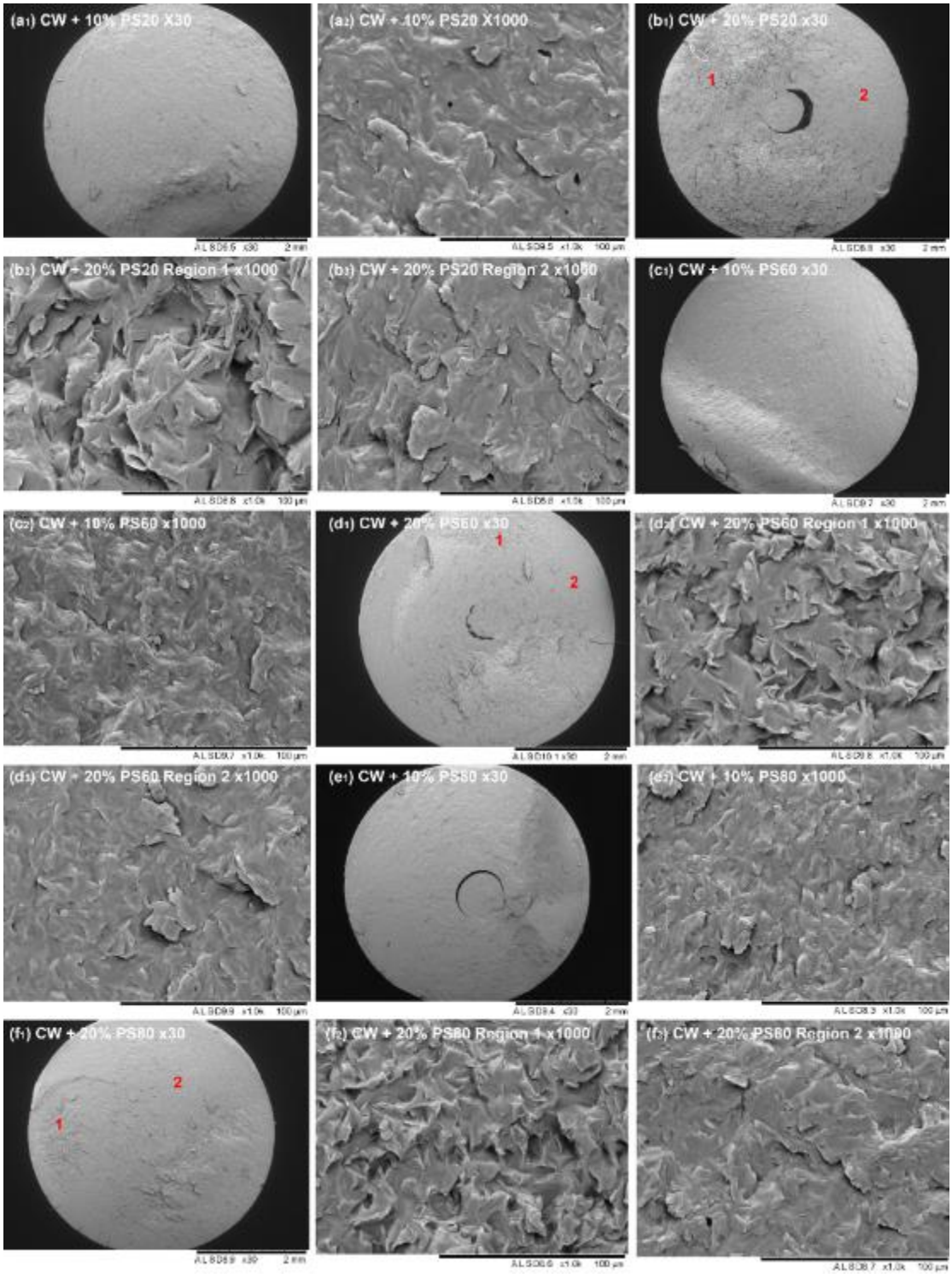


Figure 11 SEM micrographs of the fracture surfaces of specimens plasticised by polysorbate 20, 60 and 80 at various concentrations

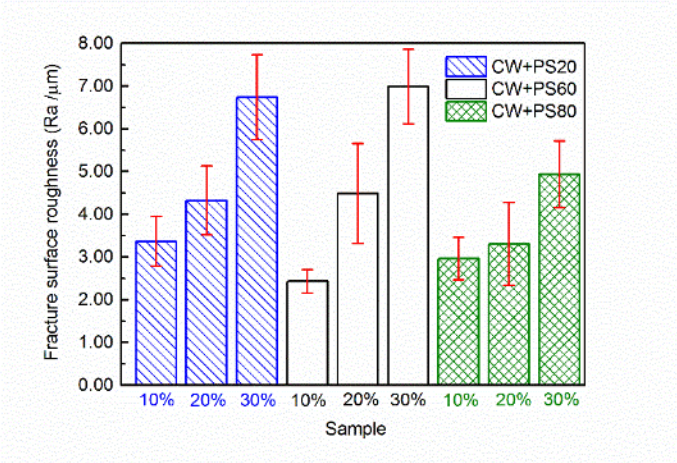


Figure 12 Fracture surface roughness (Ra) of specimens plasticised by polysorbates at different concentrations

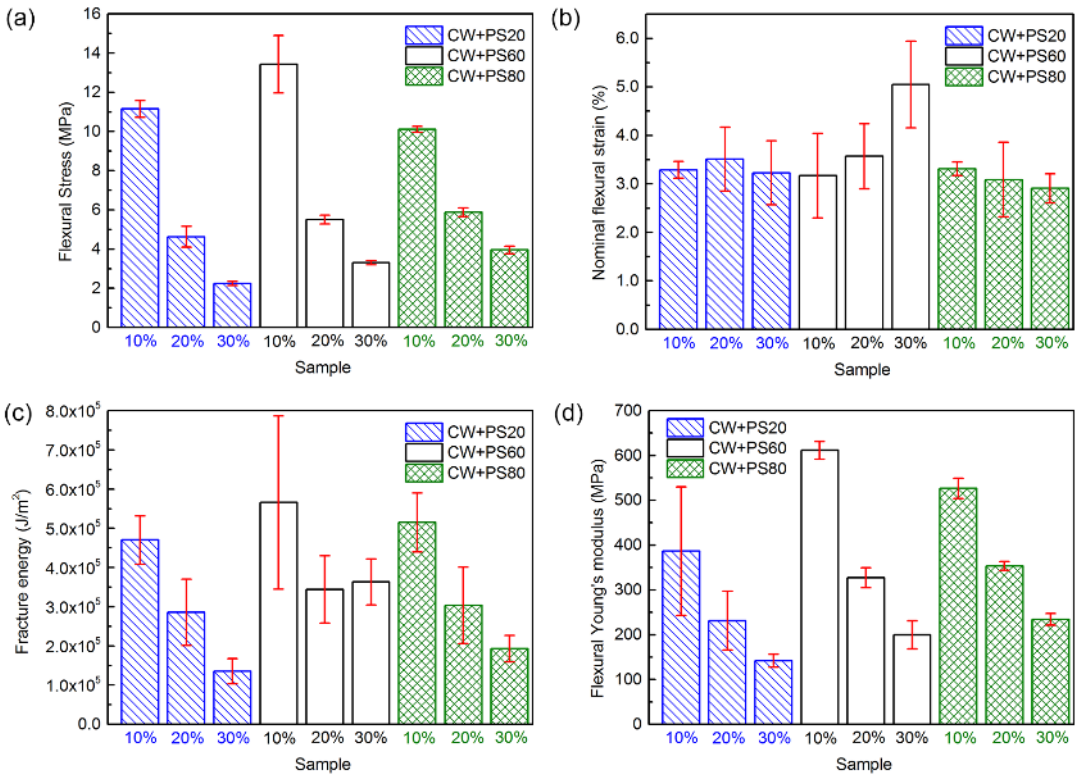


Figure 13 (a) Flexural stress (b) nominal flexural strain (c) fracture energy (d) flexural Young's modulus of specimens modified by polysorbates at different plasticiser concentrations

3.3. Modification of Candelilla wax

Polysorbate 60 was selected to determine its plasticisation efficacy on Candelilla wax at a concentration of 30 w/w%. The loading characteristics of pure and polysorbate 60 modified Candelilla wax from flexural tests are sketched in Figure 14. Although the Young's modulus and flexural load at fracture are reduced, there does not appear to be any evidence of significant plastic deformation.

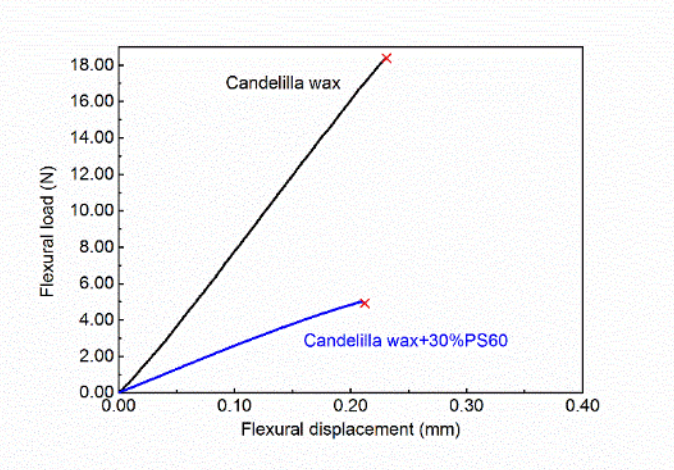


Figure 14 The loading characteristics of pure Candelilla wax and Candelilla wax + 30 w/w% polysorbate 60

3.4. Discussion

Beeswax is pliable at room temperature and has been reported to contain appreciable proportions of esters derived from hydroxy acids and diols with hydroxyl groups.[16, 17] Hydroxy acid ester has been patented to plasticise urethane elastomers.[27] Koster Keunen has also developed a synthesized Kester wax K82P containing a range of hydroxy polyesters to mimic the polyester fraction of beeswax, which is responsible for the plasticity of beeswax.[28] Therefore the hydroxyl group is postulated to play an important role in the plasticisation process, which may contribute to the improvement of cohesion due to the formation of hydrogen bonding between the plasticiser and wax molecules. The weight percentage was maintained constant in the comparison of all plasticisers used in the current work. For a unit gram of the mixture, the number of hydroxyl group (N_{hydroxyl}) contributed by the plasticiser can be denoted by:

$$N_{\text{hydroxyl}} = nN_A/M$$

where N_A is the Avogadro number, M is the molar mass of the plasticiser molecule, and n is the number of hydroxyl groups contained in each plasticiser molecule. According to Table 1, the number of hydroxyl groups in a unit gram of polysorbates is more than that in a unit gram of Span®65, which could be one factor contributing to the superior plasticising effect of polysorbates. This conclusion is further supported by the experimental data for Candelilla wax modified by polysorbate 60. The distinction between Candelilla wax and carnauba wax is a considerably greater content of hydrocarbons (C29-C33, mainly C31) in the former, which accounts for ca. 50% of the total.[28-30] The high concentration of hydrocarbons renders its molecules non-polar and there are insufficient polar groups to form intramolecular hydrogen bonds. This deficiency in cohesion between molecules, sketched as the plasticiser molecules in crystallite 1 and 2 in Figure 15, may be the fundamental reason underlying the ineffectiveness of polysorbate 60 as a plasticiser for Candelilla wax in spite of the presence of the highly branched PEGylated sorbitan group.

Waxes at the molecular level consist of at least three structurally distinctive fractions of various degrees of order and composition.[31] Aliphatic chains are assembled orderly in an orthorhombic crystal lattice at room temperature and this region is denoted as zone A in Figure 15.[32-37] Due to the polydisperse chain lengths of various molecules, the chain ends of some molecules cannot be accommodated completely within the crystalline zone A and dangle between regions of different crystalline zones accordingly. This region is termed as zone B and is in a solid amorphous state with a higher degree of mobility freedom.[31] Wax molecules excluded from the crystalline zone A such as short-chain aliphatics and cyclic compounds constitute another amorphous zone D and may also occupy zone B to some extent.[31] A further amorphous zone C has also been reported in synthetic Fischer-Tropsch waxes,[38] however it is presumed that such regions do not exist in plant waxes.[31] Consequently zone C is not sketched for the model used here. Span® 65 comprises three

flexible aliphatic chains in one molecule. If these three chains could assume completely random conformations and mix freely with carnauba wax molecules, the more branched structure would disrupt the orderly packing of wax molecules and decrease the degree of crystallinity dramatically. However, the degree of crystallinity for Span®65 plasticised wax is approximately of the same order with pure carnauba wax having a slight difference of 3.2%, as listed in Table 2, which contradicts with the above assumption.

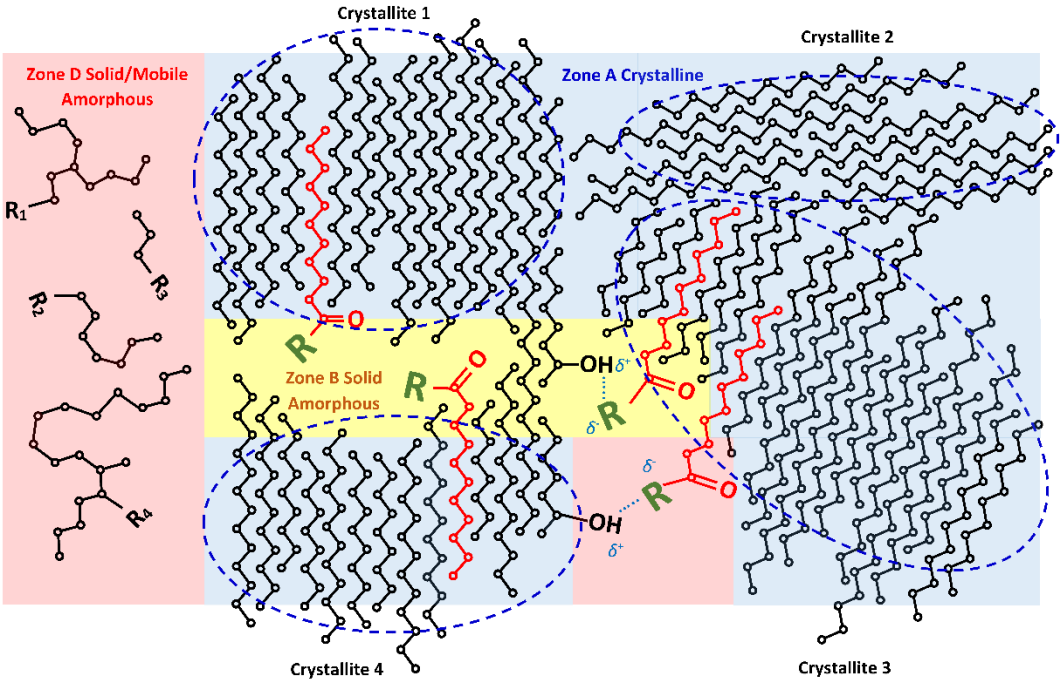


Figure 15 A schematic illustration of the molecular model of plasticised carnauba wax. Black zig-zag molecules in zone A represent aliphatic chains of carnauba wax aligned orderly to form the crystalline structure; red zig-zag molecules represent the aliphatic groups of plasticisers and R represent the branched group in plasticisers (e.g. R represents PEGylated sorbitan for polysorbates); molecules with R_1-R_4 groups in zone D represent various other molecules of carnauba wax that are not crystalline and are excluded from zone A. Each crystallite is enclosed with dashed lines. The sketch is for illustration purpose only and is not drawn to scale or represent the true chain length, structure or molecule size.

It is therefore reasonable to believe that the plasticiser molecules cannot assume completely random conformations or mix freely within carnauba wax, leading to a very low probability

of accommodating the whole plasticiser molecules completely in either zone A or zone D. Plasticiser molecules are accordingly speculated to partially participate in crystallisation with the aliphatic chains in Span®65 and polysorbates aligned in zone A as indicated by the red molecules in Figure 15. However, due to the dissimilar structure, the cyclic structures of these molecules cannot be accommodated within the crystalline zone A and thus will be located within the amorphous zone B and D. In this way, only part of the plasticiser molecules participates in an increase of the free volume in the amorphous zones, and part of them also contribute to the crystallinity increase in zone A. This may explain the limited reduction in crystallinity at a 30 w/w% polysorbate concentration. The PEGylated sorbitan in polysorbates is expected to be able to increase the free volume in the amorphous zones more effectively than the much smaller ring structure attached to the aliphatic chain in Span®65, causing a more prominent plasticisation effect. Plasticisers are capable of contributing to wax crystallisation due to the formation of their own crystals and participating in crystalline zones of individual wax molecules. The formation of their own crystals is postulated less likely for polysorbates because they are in a liquid state at room temperature, but it is possible for Span®65 due to its powder form, which is also supported by the DSC thermogram (curve 5 of Figure 8). Additionally, the net effect on the degree of crystallinity is determined by their contribution to both crystallisation and amorphisation. In this work, it is postulated that amorphisation dominates crystallisation, which contributes to the overall reduction in the degree of crystallinity of the mixtures and is in agreement with the improvement of the plasticity characteristics, as represented by the nominal flexural strain, which increases as the degree of crystallisation decreases (Figure 16).

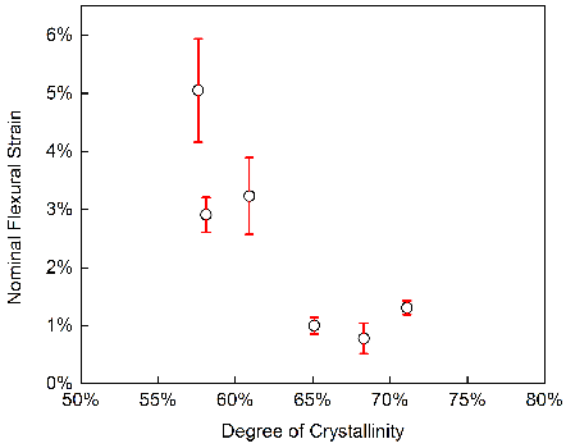


Figure 16 Nominal flexural strain as a function of the degree of crystallinity

Of all the polysorbates examined, polysorbate 60 seemed to have a superior plasticisation effect compared with polysorbate 20 and 80, on the basis of the data in Figure 1. The inferiority of polysorbate 80 is likely to be caused by the stiff double carbon-carbon bond in the aliphatic chain, which renders the molecule less mobile and flexible. This interprets the lack of an additional benefit to further increasing the concentration of polysorbate 80 above 10 w/w%. However, for polysorbate 20, it is probable that the inhomogeneity in composition due to the poor mixing behaviour caused by the smaller hydrophobicity imparted by a shorter aliphatic chain attached to the PEGylated sorbitan is the main reason for the limited sensitivity to concentration. Beeswax does not seem to influence the plasticity of the mixture substantially either. The effective molecules (including esters, free alcohol and free fatty acids with a hydroxyl group add up to 25 w/w% of beeswax based on the data presented in Table 1) and the concentration of these esters is only 7.5 w/w% in the final wax mixture which comprises 30 w/w% beeswax. The total contribution of hydroxyl groups from the beeswax to the total wax mixture may not be sufficient to produce a significant difference. In addition, 70% of the constituent hydroxy acids in beeswax is 15-hydroxyhexadecanoic acid mainly[16], which has long straight aliphatic chains contributing to the crystalline zone. This is also the case for the major diol (42.2% 1, 23-tetracosanediol, 20.2% 1, 25-hexacosanediol, 26.0% 1, 27-octacosanediol) and hydroxyl ester constituents reported [16] without

significantly branched or cyclic groups that are essential for free volume expansion. Based on the experimental results and the above analysis, it may be concluded that both hydrogen bonding and a free volume increase may be necessary for successful plasticisation of carnauba wax.

It is generally believed that effective plasticisers can usually reduce the glass transition temperature of the original polymer.[8] However, the identified effective plasticisers for carnauba wax (polysorbate 20, 60 and 80) in the current work have shown not to conform to this rule based on the DSC thermograms in Figure 8. Nonetheless, it has also been reported that the melting point is an indication of the individual crystallite size, while the degree of crystallinity represents the percentage of crystalline regions in total.[39] Therefore it is postulated that polysorbates reduce the percentage of total crystalline regions in carnauba wax while retaining the crystallite size. This explanation is also consistent with the analysis in Table 2. The calculation of the crystallite size suggested that it is of the same magnitude for all specimens. The lack of a prominent discrepancy in the peak width from the XRD diffractograms in Figure 9 also implies that the crystallite size should be similar based on the established fact that smaller crystallites produce broader peaks.[40, 41] The fatty acids with which PEGylated sorbitan is esterified to form polysorbates (lauric acid (C_{12}) for polysorbate 20, stearic acid (C_{18}) for polysorbate 60, and oleic acid (C_{18}) for polysorbate 80) have relatively much shorter chains than carnauba wax (C_{50}). The contribution of these aliphatic chains participating in the crystallisation in zone A to the crystallite size increase may not be as prominent as the contribution of the larger PEGylated sorbitan molecules involving in amorphisation to the decrease of the crystalline region percentage.

4. Conclusions

Five potential materials have been investigated in the current work to improve the plasticity of pure carnauba wax. Mechanical measurements demonstrated that beeswax and Span® 65 could not substantially reduce the brittleness of this wax. However, polysorbates could successfully impart a considerable degree of plasticity. The Young's modulus and ultimate

flexural strength were reduced significantly while the fracture energy and flexural strain increased. DSC thermograms suggested that the melting point peaks were not affected by polysorbates. XRD diffractograms revealed that the crystallinity of the wax mixtures with polysorbates were all reduced, with a maximum reduction by ~ 10% in polysorbate 60 plasticised wax while calculations using the Scherrer equation with the FWHM model suggested that the effect on the crystallite size was negligible. SEM micrographs and WLI scanning confirmed that the failure surfaces produced by polysorbates became less smooth. The effect of polysorbate concentration on plasticisation was investigated. There was not a benefit in increasing the concentration of polysorbate 20 and 80 since the nominal maximum flexural strain did not increase. This was attributed to the existence of carbon-carbon double bonds for polysorbate 80, and poor mixing behaviour of carnauba wax with polysorbate 20. A free volume mechanism increase due to the larger size of branched groups in polysorbates excluded from the crystalline zone, together with hydrogen bonding was proposed to explain the superior plasticisation effect of polysorbates compared with Span®65.

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